

CLAIMS

1. Polynucleotide molecule of 601 nucleotides isolated from *Neospora caninum* and characterised by SEQ ID NO: 9, corresponding to NcSAG4 gene, that encompasses an ORF of 522 nucleotides encoding the antigenic protein NcSAG4 of 173 amino acids and characterised by SEQ ID NO: 10.

2. Polynucleotide molecule encompassing the sequence of ORF of the NcSAG4 gene according to claim 1, included in an expression vector, and preferably plasmid pcDNA3.1-His-C (Invitrogen), by insertion of the same amplified by PCR using oligonucleotides FNcSAG4 and ReNcSAG4 characterised by SEQ ID NO: 11 and SEQ ID NO: 12, respectively.

3. Polynucleotide molecule encompassing the sequence including from nucleotide 83 to 444 of the ORF of gene NcSAG4 described in claim 1, included in an expression vector, and preferably plasmid pRSET-C, by inserting of same amplified by PCR using oligonucleotides F85NcSAG4 and Re444NcSAG4, characterised by SEQ ID NO: 13 and SEQ ID NO: 14, respectively.

4. Polynucleotide molecule encompassing the sequence including any fragment of the gene described in claim 1, or sequences obtained by any chemical, physical, enzyme or any other modification making a change in this sequence, and included in any recombinant vector.

5. The use of oligonucleotides: SAG4-2, SAG4-3, SAG4-4, 1R5SAG4, 2R5SAG4, 1F3SAG4 and 2F3SAG4, FNcSAG4, ReNcSAG4, F85NcSAG4, Re444NcSAG4 characterised by SEQ ID NO: 2, 3, 4, 5, 6, 7, 8, 11, 12, 13 and 14, for the detection of *N. caninum* by PCR or RT-PCR for use as DNA probes or for amplification by PCR of any fragment of the sequence described in claim 1.

6. A recombinant vector encompassing the nucleotide sequence characterised by SEQ ID NO: 9 according to claims 1 to 4.

7. Host eukaryote cells transfected with recombinant vectors of claim 6.

8. Host prokaryote cells transformed with the recombinant vectors of claim 6.

9. A polypeptide substantially purified or isolated selected from (a) antigenic protein NcSAG4 of *N. caninum*, characterised by SEQ ID NO: 10 according to claim 1; (b) chemical or enzyme changes of same; (c) a polypeptide consisting of a substantial portion of protein NcSAG4 of *N. caninum* or the same chemically or enzymatically modified; (d) a recombinant protein including protein or polypeptide of (a), (b) or (c).

10. The use of the promoter of gene NcSAG4 to express heterologous genes in cells of *N. caninum* transfected by gene constructions prepared with the above promoter.

11. Use of polynucleotide molecules described in claims 1 to 5 for the diagnosis of chronic infection by *N. caninum* from tissues or fluids from animals infected by PCR or RT-PCR, hybridization *in situ* with DNA probes or any other detection method based on nucleic acids of the parasite.

12. Use of the polypeptides described in claim 9 for the serological diagnosis of chronic infection by *N. caninum* by enzyme immunoassay (ELISA), radioimmunoassay (RIA), immunoblot or any other method based on the antigenicity of these polypeptides.

13. Use of monoclonal antibodies or specific polyclonal sera against the polypeptides described in claim 9, for the diagnosis of chronic infection by *N. caninum* by competition ELISA.

14. Use of monoclonal antibodies or specific polyclonal sera against the polypeptides described in claim 9, for the diagnosis of chronic infection by *N. caninum* in tissues from animals by immunohistochemistry, immunofluorescence or any other method based on the detection of the parasite by the above serum.

15. An immunogenic composition encompassing: (a) a polypeptide described in claim 9; (b) a polynucleotide molecule according to claims 1 to 4; (c) a recombinant vector as described in claim 6; (d) host cells transfected according to claim 7; or (e) host cells transformed according to claim 8, formulated as vaccine against neosporosis.

16. An immunogenic composition according to claim 15, encompassing an adjuvant or one or several cytokines.

17. A method of preparation of an immunogenic composition encompassing a combination: (a) a polypeptide according to claim 9; (b) a polynucleotide molecule containing a sequence coding the polypeptide of claim 9; (c) a recombinant vector as those described in claim 6; (d) the host cells transfected according to claim 7; (e) host cells transformed according to claim 8, formulated as vaccine against neosporosis.

18. A vaccination kit for mammals against neosporosis encompassing a container including an immunogenic composition formulated as vaccine according to claims 15, 16, and 17.